TOXICITY OF PARAQUAT TO *PARACALLIOPE FLUVIATILIS* (AMPHIPODA)

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ABSTRACT

Toxicity of the bipyridilium herbicide paraquat to the amphipod *Paracalliope fluviatilis* was investigated in the laboratory. Amphipods died in concentrations as low as 0.05 ppm. Mortality was greater among smaller amphipods. A delayed toxic effect was exhibited, with 70% of the amphipods accumulating a lethal dose after 10 h in 0.1 ppm paraquat. Paraquat adsorbed to sediment was found to be still available for uptake by amphipods, although mortality rates were lower than those in paraquat solution.

INTRODUCTION

Paraquat is a bipyridilium herbicide commonly used to control aquatic weeds. Studies of its effects on aquatic invertebrates have been carried out overseas (Newman and Way 1966, Way *et al.* 1971, Brooker and Edwards 1973), but there has been only one study in New Zealand (Burnet 1972). Burnet investigated the effects of 2 ppm paraquat applied to a Canterbury stream and found increased numbers of dead amphipods in drift samples taken on the day of application. The present study investigates this direct toxicity in laboratory experiments using the common freshwater amphipod *Paracalliope fluviatilis*.

METHODS

Methods were based on standard fish toxicity methods published by the American Public Health Association (1960). Tests were carried out in 1000 ml uncovered glass jars filled with 990 ml artesian water. The depth of the water was 120 mm and tests were carried out under static conditions with no aeration. Amphipods were collected from the Avon Stream, and ten were placed in each jar and acclimated overnight. At the beginning of each experiment the desired concentrations of paraquat were obtained by adding 10 ml paraquat solution to each jar. Three replicates were run at each concentration, and three control jars contained only artesian water. Paraquat used was the commercial formulation "Gramoxone", which contains the dichloride salt of paraquat. The stock solution was 200 000 ppm and dilutions were made with distilled water.

The experimental jars were kept at 13 ± 2°C in an enclosed wooden box, which was completely blackened inside. A 20 W fluorescent tube set along the inside back edge provided an

8h:16h light:dark cycle. The amphipods remaining alive in each jar were counted at 2 h intervals after addition of paraquat. An amphipod was considered dead if it did not swim when prodded with a blunt metal seeker. Variation between the three replicates was slight, and results were combined in all experiments to obtain percentage survival, based on 30 amphipods.

EXPERIMENT 1: EFFECT OF VARIOUS CONCENTRATIONS

Five paraquat concentrations: 1.0, 0.5, 0.1, 0.05 and 0.01 ppm were used. The number of amphipods remaining alive in each concentration was noted at intervals up to 72 h.

EXPERIMENT 2: VARIATION IN SUSCEPTIBILITY WITH SIZE

Amphipods were sorted into two size classes, large and small. These were tested in 1.0 ppm paraquat for 12 h. Dead amphipods were preserved in formalin and later measured under a binocular microscope with an eyepiece micrometer (32X magnification). The mean preserved length of the small size class was 2.0 mm, and that of the large size class 3.8 mm.

EXPERIMENT 3: EFFECT OF EXPOSURE TIME

Twenty-one jars, each containing ten amphipods, were set up at 0.1 ppm paraquat, and three jars were kept as controls. Amphipods from three of the jars were transferred to fresh water at each of 2, 4, 6, 8, 12 and 24 h after the beginning of the experiment. Survival of amphipods was determined at intervals up to 54 h.

EXPERIMENT 4: UPTAKE OF PARAQUAT FROM SEDIMENT

Stream sediment was dried and sorted according to particle size, prior to the experiment. The finest fraction less than 180 µm diameter, was used. This was a mixture of clays, silt and organic matter. Fifty g was placed in each of fifteen experimental jars, which were then filled with 950 ml of artesian water, stirred, and allowed to settle. Paraquat was added to nine of these jars, establishing three replicates at 10.0, 5.0 and 1.0 ppm. The paraquat was stirred into the water, but this disturbed the sediment very little. The remaining six jars were left as controls.

After 24 h the water was carefully decanted from each treatment. Sediment in the jars was left for a further 24 h to dry in air, so that no free water remained on the sediment surface. Fresh artesian water was then added to each jar, and left for 3 h to resettle. Ten amphipods, which had been collected the previous day, were then added to the water above the sediment in each jar. Those surviving were counted after 8, 24, 48 and 74 h. After the experiment the water above the sediment was analysed for paraquat using the method of Calderbank and Yuen (1965).

RESULTS AND DISCUSSION

EXPERIMENT 1: EFFECT OF VARIOUS CONCENTRATIONS

Paraquat is toxic to *Paracalliope*, survival times being

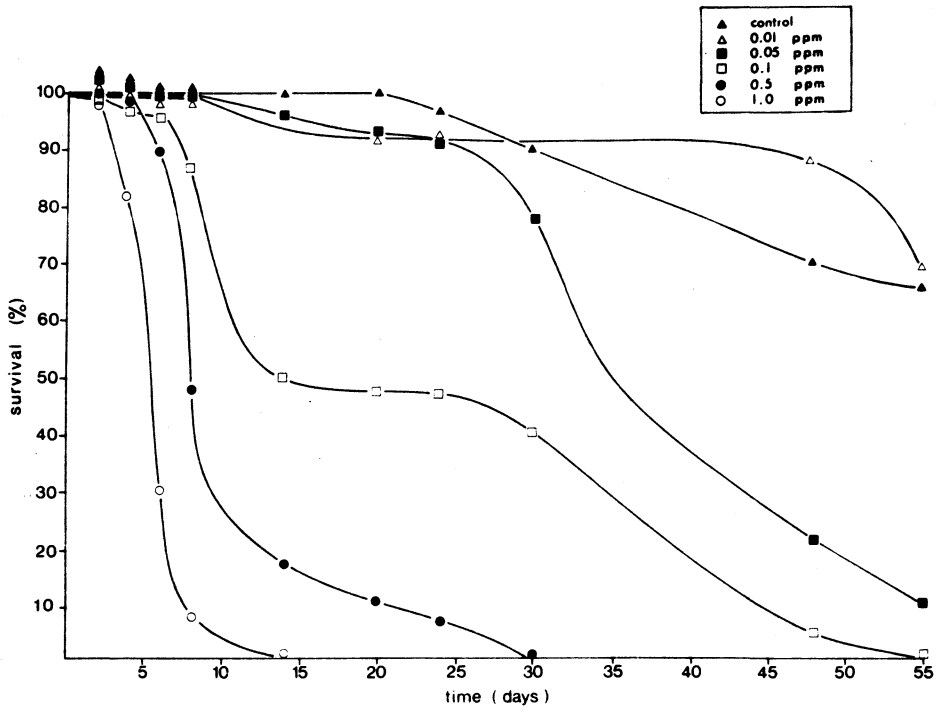

Fig. 1. Percent survival of amphipods in 6 concentrations of paraquat and in freshwater.

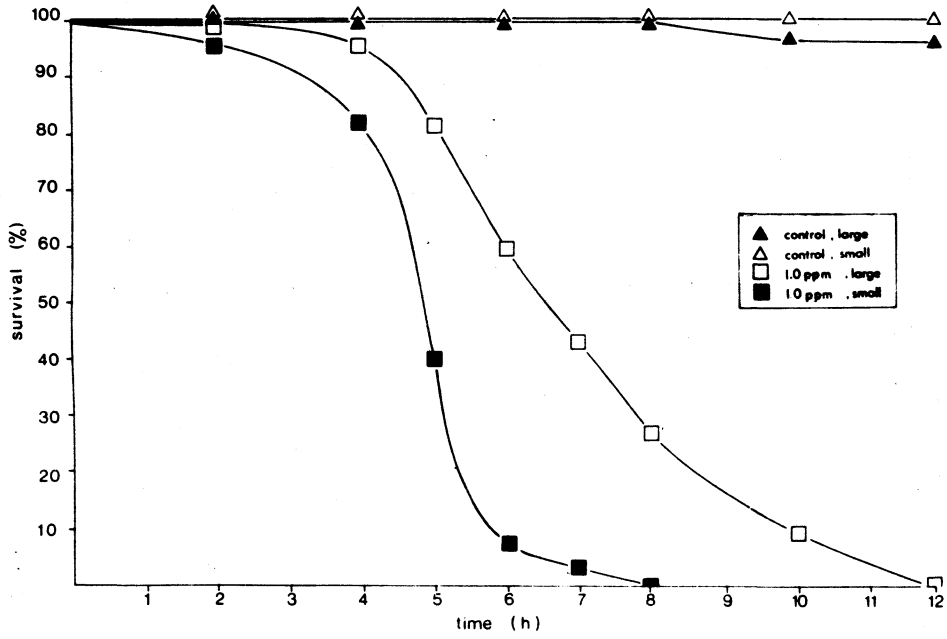

Fig. 2. Percent survival of large and small amphipods in 1.0 ppm paraquat.

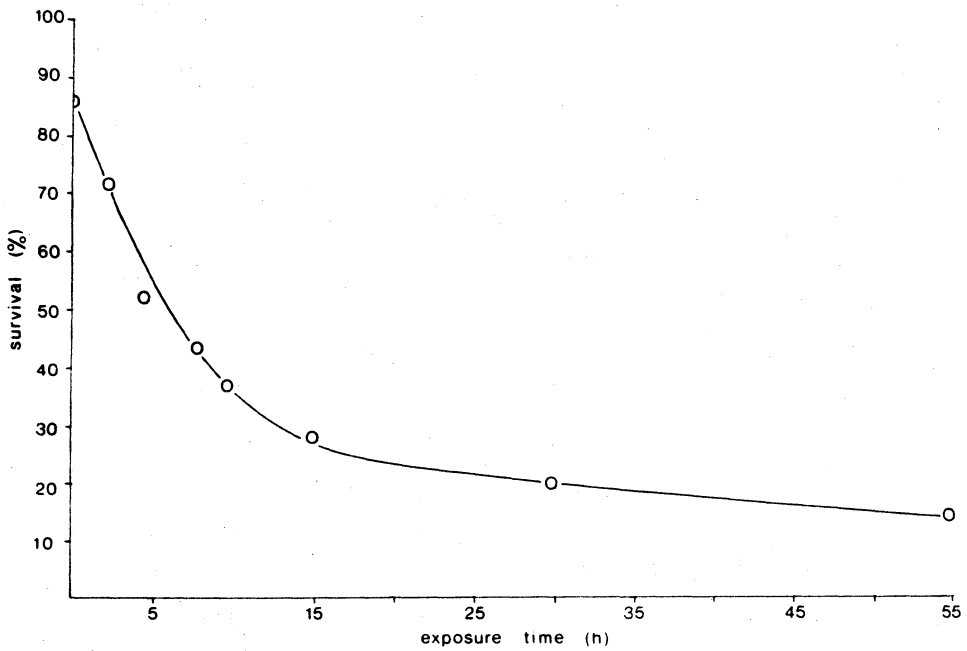

Fig. 3. Percent survival of amphipods in fresh water after different exposure times to 0.1 ppm paraquat.

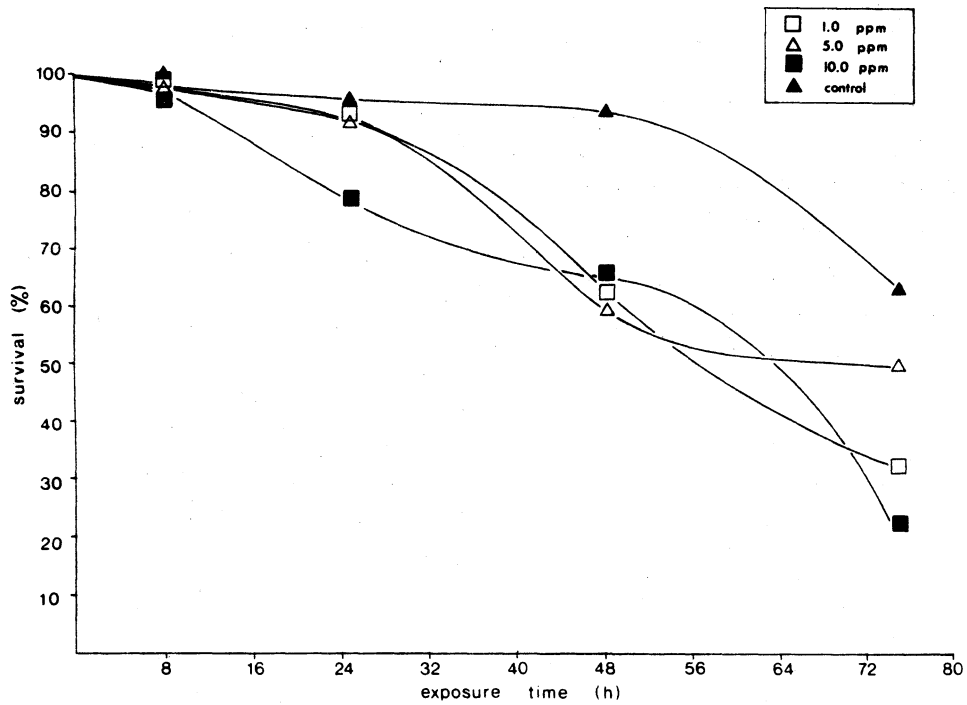

Fig. 4. Percent survival of 4 groups of amphipods exposed to sediment previously treated with different concentrations of paraquat.

inversely related to concentration (Fig. 1). The lowest concentration which produced a direct toxic effect was 0.05 ppm. This result is compatible with the results of other laboratory studies, such as that of Wilson and Bond (1969), who found that the amphipod *Hyalella azteca* was killed by 0.58 ppm of the related herbicide, diquat. Brooker and Edwards (in preparation) noted that the isopod *Asellus meridianus* died in 0.2 ppm paraquat.

EXPERIMENT 2: VARIATION IN SUSCEPTIBILITY WITH SIZE

Few laboratory studies on herbicide toxicity to invertebrates have investigated the relation between animal size and susceptibility. In the present study the susceptibility of *Paracalliope* appeared to vary with size, smaller amphipods dying before larger ones in 1.0 ppm paraquat (Fig. 2). Assuming that paraquat enters through unsclerotized regions of the amphipod cuticle, this may be attributable to a greater surface area to volume ratio in the smaller amphipods.

EXPERIMENT 3: EFFECT OF EXPOSURE TIME

The percentage survival of *Paracalliope* decreased with increased time of exposure to 0.1 ppm paraquat (Fig. 3). The lethal action of the herbicide was delayed, as shown by subsequent death of amphipods moved from paraquat solution to fresh water. This is in agreement with Crosby and Tucker (1966) who found that *Daphnia magna* individuals surviving 26 h in paraquat and then transferred to fresh water, subsequently died. In the present study most amphipods accumulated lethal doses within 10 h. Hence, although paraquat may disappear from a treated stream within several hours, there may have been sufficient time for some of the amphipod population to accumulate a lethal dose.

EXPERIMENT 4: UPTAKE OF PARAQUAT FROM SEDIMENT

Survival of amphipods exposed to sediment treated with paraquat was lower than the control in all three test treatments (Fig. 4). Analysis of the water above the sediment showed that there was no paraquat present in solution. This suggests that the amphipods may have died from the uptake of paraquat adsorbed to sediment. This could occur in the gut after sediment ingestion, or by absorption through the body surface during burrowing. Mortality rates of amphipods were lower in this experiment than in paraquat solution (Experiment 1). However, in treated streams paraquat would be present on sediment long after its disappearance from the water. Hence this source of the herbicide may prolong its toxic effect in the environment.

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